SUPPLEMENTARY MATERIAL

Deaminase-independent inhibition of HIV-1 reverse transcription by APOBEC3G

Yasumasa Iwatani^{1,2}, Denise S.B. Chan³, F. Wang⁴, Kristen Stewart Maynard⁵, Wataru Sugiura², Angela M. Gronenborn³, Ioulia Rouzina⁶, Mark C. Williams⁴, Karin Musier-Forsyth^{5,7} and Judith G. Levin^{1*}

¹Laboratory of Molecular Genetics, National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD 20892

²NAIDS Research Center, National Institute of Infectious Diseases, Tokyo 208-0013, Japan

³Department of Structural Biology, University of Pittsburgh Medical School, Pittsburgh, PA 15260

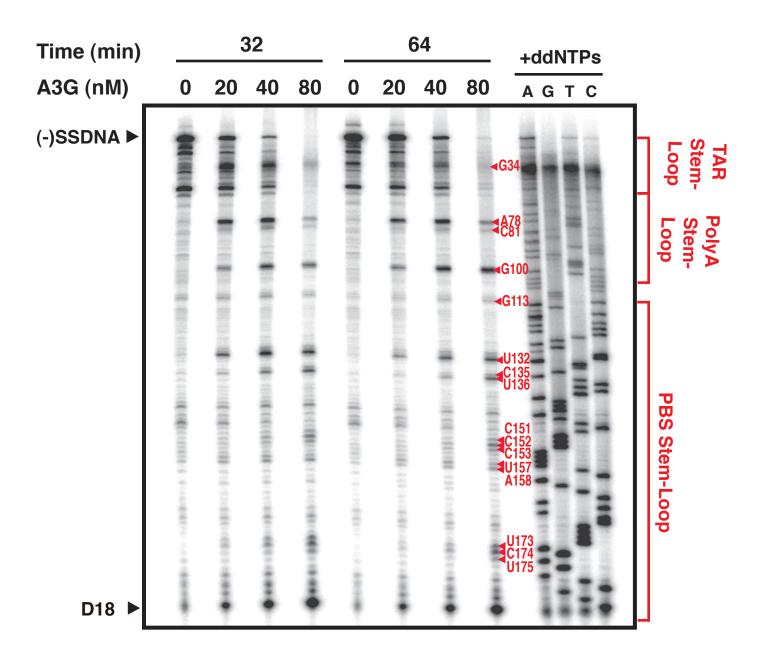
⁴Department of Physics, Northeastern University, Boston, MA 02115

⁵Department of Chemistry and ⁶Department of Biochemistry, Molecular Biology, and Biophysics, University of Minnesota, Minneapolis, MN 55455

⁷Departments of Chemistry and Biochemistry, Ohio State University, Columbus, OH 43210

^{*}Author for correspondence: levinju@mail.nih.gov

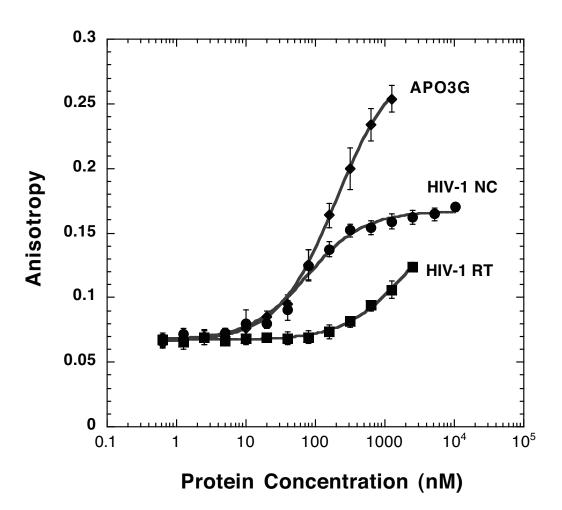
Figure S1



Legend for Supplementary Figure S1

Figure S1. Effect of increasing concentrations of A3G on (-) SSDNA synthesis primed by D18. Data are shown for reactions incubated at 32 or 64 min in the absence of A3G or in the presence of 20, 40, or 80 nM A3G. Arrows point to the positions of the (-) SSDNA product and the D18 primer. Pause sites present only in reaction mixtures containing A3G (red arrows) were mapped on the RNA 244 template (bracketed headings highlighted in red) with the help of a sequencing ladder, shown on the right. Each sequencing reaction contained only one ddNTP. The residues where the A3G-specific RT pauses are found are labeled (to the right of the gel bands). These data were used to generate the diagram that appears in Figure 3C.

Figure S2



Legend for Supplementary Figure S2

Figure S2. FA binding analysis. Various concentrations of HIV-1 NC, HIV-1 RT, or A3G were incubated with the 20-mer ssDNA, 5'-FAM-CTTCTTTGGGAGTGAATTAG-3' (JL587D). Anisotropy was plotted as a function of protein concentration in nM.